IS *KNIPOWITSCHIA CAUCASICA*-LIKE FORM FROM THE ADRIATIC SEA A NEW GOBY SPECIES? EVIDENCE FROM A MORPHOLOGICAL APPROACH IN THE EASTERN ADRIATIC SEA

by

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ABSTRACT. - Specimens of the gobiid *Knipowitschia caucasica* (Kawrajski *in* Berg, 1916) were collected since 1999 along the eastern Adriatic coast. Specimens were found in waters of different salinity, from fresh to almost sea water conditions, in springs near the sea, in small lagoons at river mouths, and in the sea-influenced parts of rivers. Standard lengths of the 87 examined specimens varied from 18.5 - 35.7 mm. The present findings confirmed previously questioned presence of this species in the Adriatic Sea and provides data on morphology and ecology of the Adriatic specimens.

RÉSUMÉ. - Est-ce que *Knipowitschia caucasica* de l'Adriatique est une nouvelle espèce de Gobiidae ? Apports de l'approche morphologique.

Des spécimens de *Knipowitschia caucasica* (Kawrajski *in* Berg, 1916) ont été collectés depuis 1999 le long de la côte adriatique orientale. Ces spécimens ont été trouvés dans des salinités et des milieux différents, allant des milieux d'eau douce aux milieux salés, ainsi que dans des deltas et des petites lagunes sous influence maritime. Les longueurs standard des 87 spécimens examinés variaient de 18,5 à 35,7 mm. Ce travail confirme la présence précédemment remise en cause de cette espèce en mer adriatique et fournit des données sur la morphologie et l'écologie des spécimens adriatiques.

Key words. - Gobiidae - *Knipowitschia caucasica* - MED - Adriatic sea - Morphology - Ecology.

The fish inhabiting transitional waters in the Adriatic Sea can be divided into two main groups: small sedentary species like Gobiidae that spend their entire life cycle there and larger migratory fishes that use those habitats for a part of their life cycle (Sparidae, Mugilidae). The first group is all-year-round completely dependable on discrete locations with transitional waters they inhabit. Therefore, they are the most vulnerable to the human threats to these locations. The conservation of these fishes depends initially on the identification of the taxon to be conserved. Therefore, the aim of the present work was to check the status of *K. caucasica*-like form in the Adriatic Sea. *Knipowitschia caucasica* (Kawrajski *in* Berg, 1916) is an euryhaline gobiid species known from the Ponto-Caspian region (Georghiev 1964, 1966; Miller, 1986) and the Mediterranean. It has been found in the Adriatic drainage basin (Miller, 1972) and the Aegean drainage basin (Economidis and Miller, 1990; Ahnelt *et al*., 1995), but its presence in the Adriatic Sea was later doubted by Economidis and Miller (1990). They believed that Adriatic population was "a *caucasica*-like form, which differs from true *caucasica* in body proportions, and may represent a new, unnamed species" (Economidis and Miller, 1990). The lateral line system, scales, coloration and fins of specimens collected since 1999 along the eastern Adriatic coast (Fig. 1) show no difference from morphological characters of *K. caucasica* described by Georghiev (1964, 1966), Economidis and

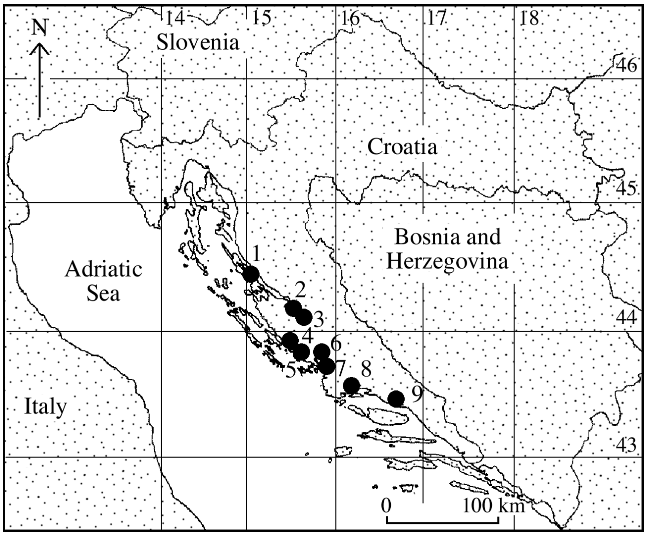

Figure 1. - Collecting sites on the Croatian coasts: (1) the island of Pag, (2) the Karin Sea, (3) river Karišnica, (4) the Vransko Lake, (5) Pirovac, (6) Prokljan lake, (7) mouth of river Jadro in the Morinj Bay, (8) spring and mouth of river Pantan in the Kaštela Bay, and (9) river Cetina.

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Miller (1990) and Ahnelt *et al*. (1995), and therefore they were identified as *K. caucasica*.

MATERIAL AND METHODS

Material

Eleven female, 26.7 + 5.6 mm to 33.8 + 5.9 mm, PMR VP1031 and 1 male, 24.5 + 4.6 mm, PMR VP1030, Jadro, Morinj Bay, Croatia, 24 Jun. 2000; 18 females, 25.9 + 5.4 mm to 35.7 + 6.7 mm, PMR VP1023 and 7 males, 24.8 + 4.9 mm to 31.1 + 5.8 mm, PMR VP1024, Karišnica, Croatia, 28 Jun. 2000; 12 females, 27.7 + 5.2 mm to 33.5 + 7.3 mm, PMR VP1036 and 11 males, 24.5 + 4.8 mm to 32.4 + 6.1 mm, PMR VP1037, Pantan, Kaštela bay, Croatia, 14 May 2001; 6 females, 27.4 + 4.4 mm to 31.2 + 5.7 mm, PMR VP1040 and 1 male, 27 + 4.7 mm, PMR VP1041, Cetina, Croatia, 16 May 2001; 16 females, 21.6 + 3.9 mm to 26.9 + 5.3 mm, PMR VP1026 and 4 males, 18.5 + 3.8 mm to 34.7 + 7.4 mm, PMR VP1027, Bilice, Prokljan lake, Croatia, 17 May 2001. Material for osteological examination: 8 females, 25.7 + 5.7 mm to 32.6 + 6.7 mm, PMR VP1034, Karin, Croatia, 28 Jun. 2000; 1 female, 31.7 + 7.4 mm, PMR VP1038 and 4 males, 29.5 + 6.8 mm to 33.2 + 7.3 mm, PMR VP1039, the mouth of the river Pantan, Split, Croatia, 14 May 2001, 4 females, 27.2 + 6.3 mm to 34.6 + 8.0 mm, PMR VP1032 and 1 male, 24.2 + 5.6 mm, PMR VP1033, the spring of the river Pantan, Split, Croatia, 14 May 2001. The specimens were deposited at the Prirodoslovni muzej Rijeka (PMR).

Methods

Morphometric and meristic methods as in Miller (1988). The morphometric data of all specimens were pooled in two groups (adult males and adult females), since even the smallest specimens (PMR VP 1026 and 1027) clearly had size and shape of urogenital papilla as adults and some of these females had ripe eggs. Fin and morphometric abbreviations are indicated in table I. The terminology of lateral-line system follows Sanzo (1911) and Economidis and Miller (1990). Head canals were examined on all 87 specimens. Distribution of rows of sensory papillae and squamation were investigated by the Iljin (1930) staining method. The six specimens from Jadro, Pantan, Cetina, Bilice and twelve from Karišnica (the total of 36 specimens) were stained and their lateral-line system and squamation examined. Vertebral counts and dorsal pterygiophores sequences were examined on eighteen cleared and stained specimens (Dingerkus and Uhler, 1977).

RESULTS

Species identification

Knipowitschia panizzae, *K. longecaudata* and *K. caucasica* differs from all other species of the genus by the following combination of characters: (1) the anterior oculoscapular and preopercular canals present, and (2) body covered with scales along lateral midline to axilla and around caudal peduncle. *K. longecaudata* is a species known from the Black Sea, Sea of Azov and Caspian Sea. The present material differs from the other Adriatic *Knipowitschia*-species, *K. panizzae* in (1) the presence of the posterior oculoscapular canal, at least in a part of population; (2) the number of vertebrae > 30, at least in a part of population and (3) the scales beginning anteriorly from rear end of D1, at least in a part of population.

Morphology

Body proportions are given in table I. Significant differences were found between sexes: males having relatively larger heads (H, Hw); longer caudal fin and bases of the first dorsal and anal fin (Cl, D1b, Ab); deeper caudal peduncle depth (CPd) and wider interorbital space (I), while females have longer distances between snout to origin of the second dorsal fin, anus and anal fin origin (SN/D2, SN/AN, SN/A), deeper body at pelvic fin origin (Vd) and longer distance from pelvic fin origin to anus (V/AN) (Fig. 2).

Fins (n = 87)

D1 V-VII (V: 5, VI: 77, VII: 5); D2 I/7-I/9 (7: 13, 8: 66, 9: 8); A I/7-I/9 (7: 16, 8: 61, 9: 10); C 11-13 branched rays (11: 26; 12: 43, 13: 18), 14-16 articulated rays (14:2, 15:82, 16:3); P 14-18 (both sides: 14 and 14: 1, 14 and 15: 3, 15 and 15: 23, 15 and 16: 16, 16 and 16: 36, 16 and 17: 4, 17 and 17: 3. 17 and 18: 1). Fin-bases and lengths in proportion to standard body length are given in table I. V rounded, anterior membrane 1/2 V I length in midline, with more or less straight rear edge. P rounded. C rounded.

Scales

Body covered with ctenoid scales, along lateral midline to axilla and around caudal peduncle. Head, breast, abdomen and anterior part of back naked. Back variably naked, scales beginning anteriorly from rear end of D1 to D2 IV-V. The scaleless area below D1 of variable size. Lower edge variably beginning anteriorly, from axilla midline to above upper corner of axilla, going straight back, and below D1 VI turning to previously mentioned rear ends. Scales in lateral series 27-37 (36 specimens, left and right side: 27: 1, 28: 2, 29: 4, 30: 10, 31: 6, 32:23, 33:11, 34: 4, 35: 6, 36: 2, 37: 3), in transverse series 7-9 (36 specimens, left and right side: 7:11, 8: 54, 9: 7).

Coloration

In life (from slides of anesthetized specimens): upper part of body dusky, with pattern of skin pigmentation brown, underside whitish to yellowish. The breast and belly

Sex	Males (n = 24)		Females (n = 63)		t value
	Range	Mean ± S.D.	Range	Mean ± S.D.	
SL (mm)	18.5 – 34.7	27.9 ± 0.4	21.6 – 35.7	28.4 ± 0.3	
%SL, H	28.1 – 31.5	29.9 ± 1.1	27.1 – 31.8	28.8 ± 1.0	4.30*
Hw	15.1 – 21.7	19.1 ± 1.7	13.8 – 21.6	17.4 ± 2.1	3.48*
SN/D1	36.3 – 41.9	38.6 ± 2.0	35.0– 43.6	39.5 ± 2.1	1.80
SN/D2	53.7 – 61.5	57.0 ± 2.0	55.4 – 62.9	59.3 ± 2.2	4.39*
SN/An	50.8 – 57.9	53.6 ± 1.5	52.1 – 59.0	55.2 ± 1.9	3.69*
SN/A	55.2 – 61.9	58.2 ± 1.6	56.4 – 64.7	60.2 ± 2.0	4.17*
SN/V	28.4 – 33.6	31.4 ± 1.7	26.7 – 35.6	30.9 ± 1.8	1.17
CP	22.6 – 27.8	25.2 ± 1.7	20.9 – 28.9	24.3 ± 1.9	2.02*
D1b	8.2 – 13.8	11.2 ± 1.8	8.2 – 13.8	10.7 ± 1.2	1.22*
D2b	14.1 – 17.7	16.0 ± 1.1	13.5 – 19.8	16.0 ± 1.3	0.23
Ab	14.3 – 17.9	16.2 ± 1.1	12.6 – 17.1	15.5 ± 1.2	2.79*
Cl	16.9 – 21.9	19.7 ± 1.6	16.0– 22.9	18.8 ± 1.8	2.23*
Pl	21.1 – 24.9	22.7 ± 1.4	20.2 – 26.7	23.0 ± 1.6	0.70
Vl	21.2 – 25.1	23.3 ± 1.2	20.1 – 26.3	23.5 ± 1.4	0.59
Vd	14.4 – 18.8	16.5 ± 1.2	14.8 – 20.9	17.5 ± 1.7	2.73*
Ad	12.4 – 17	14.6 ± 1.4	12.2 – 17.8	14.2 ± 1.5	0.86
Aw	8.1 – 12.4	10.6 ± 1.1	8.0– 13.3	10.2 ± 1.2	1.43
CPd	6.2 – 10.0	8.8 ± 0.9	5.5 – 8.6	7.1 ± 0.7	9.72*
CPw	2.7 – 5.0	4.2 ± 0.6	2.9 – 5.6	4.1 ± 0.6	0.53
V/An	21.1 – 29.0	24.4 ± 2.3	22.8 – 30.2	26.2 ± 2.1	3.53*
%CP, CPd	26.1 – 42.6	35.1 ± 4.6	22.2 – 37.1	29.3 ± 3.9	5.99*
%H, SN	17.2 – 23.5	20.6 ± 1.8	18.2 – 25.6	21.6 ± 2.1	2.27*
E	21.1 – 26.7	23.4 ± 1.4	21.1 – 27.7	23.5 ± 1.3	0.25
PO	52.6 – 58.8	56.0 ± 1.9	52.3 – 58.3	55.1 ± 1.6	2.28*
CHd	9.6 – 16.5	14.6 ± 1.8	9.1 – 16.9	14.0 ± 2.1	1.38
Hw	46.6 – 73.0	63.4 ± 6.6	49.3 –76.9	60.4 ± 6.7	1.92
%E, I	41.2 – 59.1	50.7 ± 6.1	28.6 – 52.4	37.5 ± 6.5	8.61*
%V/AN, Vl	82.4 – 107.8	95.9 ± 7.7	80.3 – 106.9	89.8 ± 6.1	3.89*

Table I. - Body proportions of *Knipowitschia caucasica*: n = number of specimens; range, mean ± S.D. = values for females and males are range, mean and standard deviation; t value = result of the t test; * = denotes significant difference ($P < 0.05$). Abbrevia-tions: Ab: anal fin base; Ad and Aw: body depth and width at anal fin origin; Cl: caudal fin length; CHd: cheek depth; CP, CPw and CPd: caudal peduncle length, width and depth, CPw measured with CPd, at minimum caudal peduncle depth; D1b and D2b: first and second dorsal fin base; E: eye diameter; H and Hw: head length and width; I: interorbital width; Pl: pectoral fin length; PO: postorbital length; SL: standard length; SN: snout length; SN/A and SN/An: distance from snout to vertical of anal fin origin and anus; SN/D1 and SN/D2: distance from snout to origin of first and second dorsal fins; SN/V: distance from snout to vertical of pelvic fin origin; V/An: distance from pelvic fin origin to anus; Vd: body depth at pelvic fin origin; Vl: pelvic fin length.

in females intensive yellow. Fins pigmentation brown, except the rear spot on D1 in males blue. The pattern of skin pigmentation is the same as in preserved specimens and it is described in detail for preserved specimens. Preserved specimens (Fig. 2): body dusky, upper part stippled, with four to five, more or less distinguished, pale saddles. Underside, including breast and belly, whitish, except dots along origin of A. Females with six to nine irregular dark blotches along lateral midline of variable size and intensity among specimens. The area around dark blotches is also stippled in some females. Head with stippled upper part, intensive preorbital bar and blotch on the chin forming with preorbital bars Y shape. D1 with one broad band in the middle of the fin; D2 and C pigmented, in some females pigment at D2 formed in rows; P with a dark mark in the origin of upper rays; A and V colourless. Males with vertical dark bars (two to three below D1, two to three below D2, none to two on caudal part) along lateral midline and dark blotches on caudal part. Some of the vertical bars below D1 and D2 reach dorsal midline in the most of the males. The area between vertical bars is also stippled. Head densely pigmented, including underside. D1 with three longitudinal bands and intensive spot in the rear part; D2 with three to

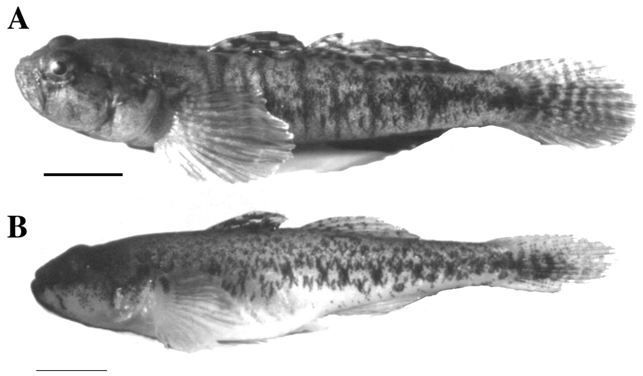

Figure 2. - *Knipowitschia caucasica*, **A**: male, 29.5 + 6.8 mm. **B**: female, 31.7 + 7.4 mm, Pantan, Kaštela bay, Croatia, 14 May 2001. Scale bar = 5 mm.

four bands, C, A, P and V pigmented. P with dark mark in origin of upper rays. Juveniles with upper part of body and origin of A pigmented. Five to nine marks along lateral midline, posteriorly joined in a longitudinal band. Preorbital bar and chin also pigmented, C dotted more intensively, D1, D2 and P poorly. The mark in the upper pectoral corner also visible.

Vertebrae and dorsal pterygiophores (n = 18)

Number of vertebrae 29-31 (29: 1; 30: 9; 31:8), including urostyle. Dorsal pterygiophore formula 3-1221001 and 3-1220101 (3-1221001: 8; 3-1220101: 5; 3-1121101: 1; 3-1131000: 1; 3-1131101: 1; 3-1211001: 1; 3-1222001: 1).

Lateral line system

Head canals (n = 87) variously developed (Fig. 3). The anterior oculoscapular and preopercular canals always present, carrying pores λ, κ, α, ρ, and γ, ε. The posterior oculoscapular canal carrying pores ρ^1 and ρ^2 absent (45 specimens) or present (42 specimens). Rows and the number of sensory papillae (n = 36) as follows (i) *preorbital*: snout with three rows in median preorbital series: internal row *r* (3-7), outer row *s* (*3-8*), anterior row s^3 (*3-6*). Lateral series *c* in four parts: superior (c^2) in the angle between AN and PN (*2-6*); middle c^1 (*2-6*) close to AN; inferior upper c_2 (*2-6*) and lower c_1 (*3-7*) above lips. (ii) *suborbital*: infraorbital row *a* variably extending forwards, from anterior to posterior edge of pupil, consisting of 6-11 longitudinally arranged papillae and a longer transverse row *atp* (*3-6*). The transverse proliferation of row *a* varying from complete absence of transverse proliferation (except *atp*) to 6 transversely doubled or tripled papillae. Longitudinal row *b* (*8-15*) anteriorly beginning below rear border of eye, occasionally extending before rear border of eye. Six to seven transverse row *c* below level of row *b*: *c1* (*2-4*), *c2* (*2-5*), *c3* (*2-6*), *c4* (*2-5*), *c5* (*2-6*), *c6* (absent or *3-5*) and *cp* (7-15). Transverse row *cp* ending below level of row *d*, rarely on the level of row *d* and occasionally *c3* crossing over row *d* and ending below its level. Longitudinal row *d* (*8-25*) continuous or with separated supralabial and posterior part, not reaching posteriorly row *cp*. (iii) *preoperculo-mandibular*: external row *e* and internal row *i* divided into anterior (*e: 15-24*, *i: 15-23*), and posterior sections (*e*: *18-27*, *i: 15-25*); mental row *f* (*3-4*). (iv) *oculoscapular:* anterior transverse series *tra* (*2-6*) behind pore α, anterior longitudinal row x^1 divided by posterior transverse row, *trp* (*5-13*) in anterior section (*8-14*) above row *z* and posterior section (*2-6*) above row *q*, posterior longitudinal row x^2 (*5-9*) over row *y* (1); row *z* (*5-13*) ends behind pore γ, row *q* (1-2) behind transverse row *trp* and below posterior section of x^1. Axillary rows as^1 (*7-11*), as^2 (*7-15*), as^3 (*4-10*), la^1 (*3-6*) and la^2 (*3-6*) present. (v) *opercular*: transverse row *ot* (*12-25*); superior longitudinal row *os* (*6-15*); and inferior longitudinal row *oi* (7-13). (VI) *anterior dorsal*: row *n* (*8-16*), row *g* (*6-13*), row *m* (*3-7*) and row *h* (*8-20*) present, row *o* absent.

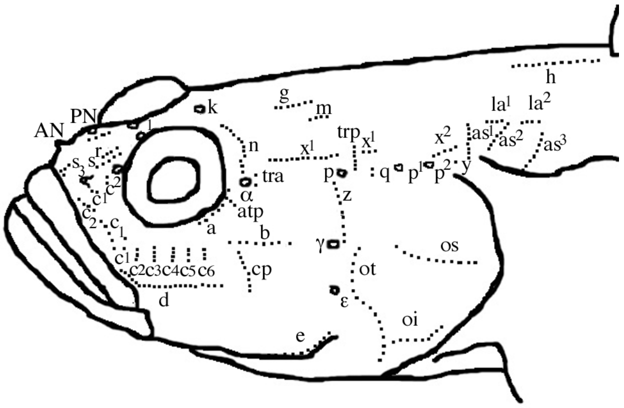

Figure 3. - *Knipowitschia caucasica*, head lateral-line sensory papillae and canal pores, female, 30.9 + 5.7 mm, PMR VP1040, Cetina, Croatia, 16 May 2001. AN, PN: Anterior and posterior nostrils; see other terminology in the text..

Ecology

The specimens were found in different salinity, from fresh to almost sea water conditions (0-24.5 PSU), in springs near the sea, in small lagoons at river mouths, and in the sea-influenced parts of rivers. The bottoms were usually a combination of bare surface area and an area covered with vegetation. The bottom structure highly varied: mud with rare gravel or individual boulders, coarse sands between large rocks, coarse gravel and cobbles mixed with bedrock, boulders and bedrock with rare patches of gravel. Vegetation varied from euryhaline sea species (*Cladophora* sp., *Chaetomorpha* sp., *Enteromorpha intestinalis, Ulva lactuca, Zostera noltii*) to freshwater species (*Chaetophora* sp., *Chara* sp., *Potamogeton natans, Potamogeton* sp., *Ranunculus trichophyllus*).

Geographical distribution

The localities listed in Material, and all the other localities where the species was found and collected are situated in northern and middle Dalmatia, Croatia: the island of Pag, the river Karišnica, the Karin Sea, the Vransko Lake, Pirovac, the mouth of the river Jadro in the Morinj Bay, the Prokljan lake, the spring and the mouth of river Pantan in the Bay of Kaštela, the river Cetina (Fig. 1).

DISCUSSION

Taxonomic characters in zoology include various kinds of characters: morphological, physiological, molecular, behavioural, ecological and geographic. Morphological and molecular characters are almost the only commonly used characters in today's gobiology. Molecular characters have become important in macrotaxonomy of this large and complex taxon in the last decades. However, on microtaxonomical level, recent descriptions of new species of European gobies still consist of morphological characters supported by a few ecological data. Among morphological data, the qualitative characters (usually coloration, lateral line system) and quantitative characters with ranges that do not overlap between taxa (usually fin and scale meristics) are used for diagnosis and differential diagnosis. For those quantitative characters with statistically significant difference between taxa, although containing ranges that overlap between taxa (usually morphometrics) there are no published examples of use for the identification of European gobies.

The present results show high variability of quantitative and qualitative morphological data between specimens of the Adriatic population of *K. caucasica*. The differences are present within samples from every locality of collecting, and the ranges of quantitative data and the character states of the qualitative data greatly overlap between localities. *K. caucasica* is a gobiid species with high intraspecific variability in important morphological characters (especially lateral line canals and coloration) (Georghiev, 1964, 1966; Economidis and Miller, 1990; Ahnelt *et al*., 1995; present data). Aside the sexual dimorphism, the polymorphism within the same sex is present between populations, but also between morphs within the same population (Ahnelt *et al*., 1995; present results). The variability of characters is geographically erratic except that the population of the Aegen Anatolian area shows higher coloration variability and additional reduction of lateral line canals in a part of population (Ahnelt *et al*., 1995). The Adriatic specimens have lower vertebral number compared to data in Economidis and Miller (1990) and Georghiev (1964, 1966). At the present state of knowledge (Georghiev, 1964, 1966; Economidis and Miller, 1990; Ahnelt *et al*., 1995; present data), intrapopulation variability of characters, as well as geographically irregular differences between populations prevent real recognition of different taxa within the species distribution.

Nevertheless, the present work proved the presence of *K. caucasica* in the Adriatic Sea, despite previous doubts (Economidis and Miller, 1990). Economidis and Miller (1990) regarded records of *K. caucasica* from Venice and Trieste (Miller, 1972) as new, unnamed species, similar to *K. caucasica*, but different in body proportions. We consider only the statistically significant differences in body proportions between populations as insufficient evidence for recognition of separate species (Mayr and Ashlock, 1991, p. 96: "If a (statistical) difference is established, additional evidence is required before it can be decided whether the populations should be considered different taxa and, if so, whether they should be ranked as subspecies or species"). All morphological characters (fins, scales, coloration, vertebrae and dorsal pterygiophores, lateral line system) of the Adriatic population we observed, important for the diagnosis of gobiid species, fit well within the previous data on *K. caucasica* (Georghiev, 1964, 1966; Economidis and Miller, 1990; Ahnelt *et al*., 1995), but in a future search for different taxa within this widespread polymorphic species the new evidences are needed from molecular, ethological or ecological characters to clarify the status of *K. caucasica* (Mayr and Ashlock, 1991, p. 92: "Sibling species can be discovered because they differ in various other attributes even when they are extremely similar in the morphological characteristics normally employed in taxonomic analysis").

The other Adriatic euryhaline species of genus *Knipowitschia*, *K. panizzae* (Verga, 1841) was redescribed by Miller (1972). The collected specimens in the present work could not be assigned to *K. panizzae* (see Specific identification). On the other hand, Miller's (1972) description of *K. panizzae* fit well within morphological characters of *K. caucasica* in view of the present state of knowledge (Economidis and Miller, 1990; Ahnelt *et al*., 1995; present data). The status of *K. panizzae* as valid species and previously recognized differing characters (squamation, modal vertebral number, anterior vertical band in males) (Miller, 1972) could be checked on a larger sample of *K. panizzae* from the type locality, Comacchio, Italy. The other findings of *K. panizzae* on the Adriatic locations mentioned in Miller (1972), Bianco and Miller (1990), Economidis and Miller (1990) become uncertain without reliable differing characters for this species. If *K. panizzae* would become part of the Adriatic population of *K. caucasica*, this would have the practical nomenclature consequence of *panizzae* Verga, 1841 becoming a senior synonym for *caucasicus* Kawrajski *in* Berg, 1916, which was pointed out already by Miller (1972).

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